CALIFORNIA ENVIRONMENTAL PROTECTION AGENCY
DEPARTMENT OF PESTICIDE REGULATION
MEDICAL TOXICOLOGY BRANCH

SUMMARY OF TOXICOLOGY DATA

Metofluthrin

Chemical Code # 5943, **Tolerance #** 53008
SB 950 # New A.I.

11/17/06

I. DATA GAP STATUS

Chronic toxicity, rat:	No data gap, no adverse effect evident
Chronic toxicity, dog:	No data gap, possible adverse effect
Oncogenicity, rat:	No data gap, possible adverse effect
Oncogenicity, mouse:	No data gap, no adverse effect evident
Reproduction, rat:	No data gap, no adverse effect evident
Teratology, rat:	No data gap, no adverse effect evident
Teratology, rabbit:	No data gap, no adverse effect evident
Gene mutation:	No data gap, no adverse effect evident
Chromosome effects:	No data gap, no adverse effect evident
DNA damage:	No data gap, no adverse effect evident
Neurotoxicity:	No data gap, possible adverse effect

Toxicology one-liners are attached.

All record numbers through 225391 were examined.
** indicates an acceptable study.
Bold face indicates a possible adverse effect.
indicates a study on file but not yet reviewed.
File name: T061117
Revised by T. Moore, 11/17/06

II. TOXICOLOGY ONE-LINERS AND CONCLUSIONS

These pages contain summaries only. Individual worksheets may contain additional effects.

COMBINED, RAT

**** 0065; 225374**; "Combined Chronic Toxicity/Oncogenicity (Feeding) Study in the Rat"; (H. Schmid, D. Flade, P. Gretener, K. Weber; RCC Ltd., Toxicology, CH-4452 Itingen, Switzerland; Study No. 846244; 7/7/05); Fifty HanBrl:WIST (SPF) rats/sex/group were treated in the diet with 0, 20, 200, 900 or 1800 ppm of S-1264 (lot no. PK-020301 G, purity: 96.6%) for 24 months ((M) 0, 0.84, 8.24, 38.1, 77.8 mg/kg/day, (F) 0, 1.03, 10.1, 47.4, 96.1 mg/kg/day). An additional 20 animals/sex/group received the test material in the diet for 12 months. Survival of the study animals was not affected by the treatment. The mean body weights of both sexes in the 1800 ppm group and the males in the 900 ppm group were less than those of the controls ($p<0.01$ or 0.05). The mean food consumption of both sexes in the 1800 ppm group and the males in the 900 ppm group was less than that of the controls, particularly during the first nine months of the study ($p<0.01$ or 0.05). No treatment-related effects were evident in the hematology, urinalysis or ophthalmological data. In the clinical chemistry evaluation, the mean serum cholesterol and phospholipid levels of the 900 and 1800 ppm males were greater than those of the control over the course of the 12 month evaluation period ($p<0.01$ or 0.05). Gamma-glutamyl peptidase activity was noted in the serum after 26 and 52 weeks of treatment ($p<0.01$). The concentrations of the various ions in the serum were statistically different from the control values over the course of the study. However, there was no consistent dose-response demonstrated by these data. The total protein and albumin concentrations in the serum of the 900 and 1800 ppm males and the 1800 ppm females were greater than the control values over the course of the study ($p<0.01$ or NS). The mean absolute and relative liver weights for both sexes in the 1800 ppm group were greater than the control values after 52 weeks of treatment ($p<0.01$). The mean relative liver weight of the 900 ppm males was greater, as well ($p<0.01$). After 104 weeks of treatment, the mean relative liver weights of both sexes in the 1800 ppm group and the females in the 900 ppm group were greater than the control values ($p<0.01$). In the histopathological examination, an increased incidence of hepatocellular hypertrophy was noted for the 900 and 1800 ppm females after 52 weeks of treatment and for the 1800 ppm males after 104 weeks of treatment ((M) 0: 4/50 vs. 1800: 13/50 ($p<0.01$), (F) 0: 1/20 vs. 900: 7/20, 1800: 8/20, ($p<0.01$)). The 1800 ppm males and females demonstrated an increased incidence of eosinophilic foci and mixed cell foci, respectively, in the liver after 104 weeks of treatment ((M) 0: 1/50 vs. 1800: 10/50 ($p<0.01$), (F) 0: 1/49 vs. 1800: 12/50 ($p<0.01$)). Tubular vacuolation was noted in the kidneys of the 1800 ppm males after 104 weeks of treatment (0: 0/49 vs. 1800: 8/50 ($p<0.01$). There was a dose-related increase in hepatocellular adenomas and carcinomas in both sexes of the 1800 ppm treatment groups (adenomas, (M) 0: 1/50 vs. 1800: 6/50, (F) 0: 1/49 vs. 1800: 7/49 ($p<0.01$), carcinomas, (M) 0: 0/50 vs.1800: 8/50, (F) 0: 0/49 vs. 7/49 ($p<0.01$)). **Possible adverse effect:** liver oncogenicity; **Chronic Dietary NOEL:** 200 ppm ((M) 8.24 mg/kg/day, (F) 10.1 mg/kg/day) (based upon the increased cholesterol and phospholipid levels in the serum of the 900 ppm males, the increased relative liver weights of the 900 ppm males after 52 weeks of treatment and the 900 ppm females after 104 weeks of treatment and the increased incidence of hepatocellular hypertrophy in the 900 ppm females after 52 weeks of treatment); **Oncogenicity evident. Study acceptable.** (Moore, 8/25/06)

CHRONIC TOXICITY, RAT

See Combined, Rat above.

CHRONIC TOXICITY, DOG

**** 0062; 225371**; "12-Month Repeated Dose Oral Toxicity Study of S-1264 in Dogs"; (H. Uchida; Panapharm Laboratories Co., Ltd., Uto-shi, Kumamoto 869-0425, Japan; Study No. P020637; 5/14/04); Four beagle dogs/sex/group were dosed orally in capsules with 0, 10, 30 or 100 mg/kg/day of S-1264 (lot no. PK-020301 G, purity: 96.6%) for 12 months. No deaths resulted from the treatment. The males in the 30 and 100 mg/kg groups and the females in the 100 mg/kg group exhibited tremors through out the study. There was an increased incidence of emesis for the 30 mg/kg males and for both sexes in the 100 mg/kg group as well. There was no treatment-

related effect upon the mean body weights or the food consumption. Urinalysis, hematology, clinical chemistry and ophthalmology evaluations did not reveal any treatment-related effects. No treatment-related lesions were noted in the necropsy and histopathology examinations. **Possible adverse effect:** tremors. **Chronic Oral Toxicity NOEL:** (M) 10 mg/kg/day (based upon the incidence of tremors for the males in the 30 mg/kg group), (F) 30 mg/kg/day (based upon the incidence of tremors for the females in the 100 mg/kg group); **Study acceptable.** (Moore, 8/14/06)

ONCOGENICITY, RAT

See Combined, Rat above.

ONCOGENICITY, MOUSE

** 0064; 225373; "78-Week Oncogenicity (Feeding) Study in the CD-1 Mouse"; (H. Schmid, D. Flade, P. Gretener, K. Weber; RCC Ltd., Toxicology, CH-4452 Itingen, Switzerland; Study No. 847663; 7/7/05); Fifty two CD-1 mice/sex/group were dosed in the diet with 0, 100, 1000 or 1750 ppm of S-1264 (lot no. PK-020301 G; purity: 96.6%) for 78 weeks ((M) 0, 11.77, 115.7 and 208.7 mg/kg/day, (F) 0, 15.40, 154.7, 276.7 mg/kg/day). An additional 12 animals/sex/group were dosed in the same manner for 52 weeks. Initially the high dose level was 2500 ppm. Twelve males and 4 females in the 2500 ppm group were found dead on day 2. The dose was adjusted to 1750 ppm from that time point (2^{nd} week) to the termination of the study. No treatment-related effect upon survival was evident once the high dose level had been adjusted. No treatment-related clinical signs were evident throughout the study. Although the mean food consumption for all of the treatment groups was lower throughout much of the study ($p<0.01$ or 0.05), the mean body weights of the treated groups were not affected. The hematology evaluation did not reveal any treatment-related effects. The mean absolute and relative liver weights for the 1750 ppm females after 78 weeks of treatment were greater than those values for the controls ($p<0.01$ or 0.05). In the histopathology examination, the incidence of hepatocellular hypertrophy in the liver was greater for both sexes in the 1750 ppm group after 78 weeks of treatment than for the controls (NS). **No adverse effect indicated. Chronic Dietary NOEL:** (M/F) 1000 ppm ((M) 115.7 mg/kg/day, (F) 154.7 mg/kg/day) (based upon the increased incidence of hepatocellular hypertrophy in the livers of the 1750 ppm treatment group); **no oncogenicity evident. Study acceptable.** (Moore, 8/23/06)

REPRODUCTION, RAT

** 0063; 225372; "Oral (Diet) Two-Generation (One Litter per Generation) Reproduction Study of S-1264 in Rats"; (A. M. Hoberman; CR-DDS Argus Division, Horsham, PA, Charles River DDS-Worcester, Worcester, MA, Research Pathology Services, Inc., New Britain, PA; Project ID No. 1119-031; 2/16/05); Thirty Crl:CD(SD)IGS BR VAF/Plus rats/sex/group were dosed in the diet with 0, 50, 200, 1000 or 1800 ppm of S-1264 (lot no. PK-020301 G; purity: 96.6%) for two generations. The treatment period for the P parents included at least 70 days prior to mating, the mating period, 3 weeks of gestation and 4 weeks of lactation. At that time, 30 F1 animals/sex/group were selected as parents and treated for a minimum of 70 days in the premating period, the mating period, and 3 and 4 weeks for the gestation and lactation periods, respectively. No parental animals died as a result of the treatment. The dams in the 1800 ppm group of both generations and the dams in the 1000 ppm group in the F1 generation demonstrated treatment-related twitches and tremors during the lactation period ($p<0.01$ or 0.05). During the premating period, the mean body weights of both sexes in the 1800 ppm treatment group, F1 generation, and the females in the 1000 ppm treatment group, F1 generation, were less than the control values ($p<0.01$ or 0.05). The lower mean body weights persisted for the females in these two groups through the end of the lactation period ($p<0.1$ or 0.05, NS). Mean food consumption for the 1000 and 1800 ppm females, F1 generation, was less than that of the controls during the premating, gestation, and lactation periods ($p<0.01$, 0.5 or NS) as well. The 1000 and 1800 ppm males in the P generation and both sexes in the 1000 and 1800 ppm treatment groups of the F1 generation had higher mean absolute and relative liver weights than did the controls in both generations ($p<0.01$ or 0.05). The mean relative liver weight of the 1800 ppm females in the P generation was greater than that of the controls as well ($p<0.05$). Hepatocellular hypertrophy was noted in the livers of the 1000 and 1800 ppm males of both

generations and the 1800 ppm females of the P generation ((M) P generation, 0: 0/30 vs. 1000: 12/30, 1800: 16/30; F1 generation, 0: 2/29 vs. 1000: 11/30, 1800: 17/30; (F) P generation, 0: 0/30 vs. 1800: 13/30). The histopathological examination did not reveal any treatment-related lesions in the reproductive organs. There was no treatment-related effect upon the reproductive parameters. The pup weaning index was reduced for the 1800 ppm pups of the F1 generation ($p<0.01$) This effect was not noted for the F2 generation. The mean pup weights for the 1800 ppm group in both generations were less than those of the control by day 21 post-partum ($p<0.01$ or 0.05). **No adverse reproductive effects indicated. Parental NOEL:** 200 ppm (based upon the hepatocellular hypertrophy noted in the liver of the 1000 ppm males of both generations and the incidence of clinical signs and lower mean body weights and food consumption noted for the females in the1000 ppm treatment group of F1 generation) ((M) 11.2 to 30.7 mg/kg/day, (F) 11.0 to 37.4 mg/kg/day), **Reproductive NOEL:** 1800 ppm (based upon the lack of treatment-related effects upon the highest treatment level) ((M) 98.8 to 310.4 mg/kg/day, (F) 96.2 to 339.7 mg/kg/day), **Developmental NOEL:** 1000 ppm (based upon lower mean pup weights for the 1800 ppm F1 and F2 pups) ((M) 53.2 to 160.3 mg/kg/day, (F): 54.3 to 201.0 mg/kg/day); **Study acceptable.** (Moore, 8/22/06)

0059; 225368; "Study of Fertility and Early Embryonic Development to Implantation of S-1264 Administered Orally to Rats"; (H. Hara, S. Suyama, T. Ushimaru, M. Kamijima; Ina Research Inc., Ina-shi, Nagano-ken 399-4501, Japan; Study No. ST01083; 6/14/02); Twenty Crj:CD (SD) rats/sex/group (unless otherwise noted) were dosed orally by gavage with 0 (vehicle: corn oil), 5 (M only), 10, 20, or 40 (F only) mg/kg of S-1264 (lot no. PK-010501 G, purity: 95.1%) for a 2-week premating period and a mating period. The males were dosed through to the time of termination, day 57, while the females were treated through day 7 of gestation. Five females in the 40 mg/kg group died, two during the mating period and three on days 5-6 of the gestation period. The females in the 40 mg/kg group exhibited tremors and salivation. There was no treatment-related effect upon the mean body weights or food consumption of the males. The females in the 40 mg/kg group demonstrated reduced body weight gain and mean food consumption during the first 7 days of gestation. Treatment did not affect the any of the reproduction parameters or the early development of the embryos. **No adverse effect indicated. Parental NOEL:** (M/F) 20 mg/kg/day (based upon the treatment-related effects noted for the females in the 40 mg/kg group and the lack of any treatment-related effects for the males in the 20 mg/kg group); **Reproduction/Early Developmental NOEL**: 40 mg/kg/day (based upon the lack of a treatment-related effect on the fetuses in the 40 mg/kg group); **Study supplemental** (non-guideline study). (Moore, 8/9/06)

TERATOLOGY, RAT

** 0058; 225367; "Study of Effects on Embryo-Fetal Development of S1264 Administered Orally to Rats"; (H. Hara, S. Suyama, T. Ushimaru, M. Kamijima; Ina Research Inc., Ina-shi, Nagano-ken 399-4501, Japan; Study No. ST01085; 4/26/02); Twenty four mated females Crj:CD (SD) rats/group were dosed orally by gavage with 0 (vehicle: corn oil), 5, 15, 30 mg/kg/day of S-1264 (lot no. PK-010501 G, purity: 94.9%) from day 6 through day 19 of gestation. All of the dams survived the treatment. Five dams in the 30 mg/kg group demonstrated tremors one or more times during the treatment period. There was no treatment-related effect on the mean body gain or food consumption during the treatment period. The treatment did not affect the development of the fetuses. There was no treatment-related incidence of anomalies in the fetuses. **No adverse effect indicated. Maternal NOEL:** 15 mg/kg/day (based upon the incidence of tremors in the 30 mg/kg treatment group); **Developmental NOEL:** 30 mg/kg/day (based upon the lack of an effect in the 30 mg/kg treatment group). **Study acceptable.** (Moore, 8/7/06)

0060; 225369; "Study for Effects on Pre- and Postnatal Development, Including Maternal Function, of S-1264 Administered Orally to Rats"; (H. Hara, S. Suyama, T. Ushimaru, M. Kamijima; Ina Research Inc., Ina-shi, Nagano-ken 399-4501, Japan; Study No. ST01084; 8/1/0); Twenty four mated female Crj:CD (SD) rats/group were dosed orally by gavage with 0 (vehicle: corn oil), 5, 15, or 30 mg/kg/day of S-1264 (lot no. PK-010501 G, purity: 94.9%) from day 6 of gestation through day 20 of lactation. The two F1 offspring/sex/litter/group were retained after the lactation period. One animal/sex/litter/group was evaluated in a behavioral test protocol through

10 weeks. The other second animal/sex/litter/group was maintained through a 10 week premating period and mated. The F1 females were subjected to a caesarean section on day 14 of gestation and the embryos examined. The parental dams in the 30 mg/kg group demonstrated tremors during the gestation and lactation treatment periods. Otherwise, there was no treatment-related effect on the dams and no effect upon the development of the F1 generation. There was no treatment-related effect upon the reproductive performance of the F1 generation. The development of the F2 generation embryos was not apparently affected through the 14th day of gestation. **No adverse effect indicated**. **Maternal NOEL:** 15 mg/kg/day (based upon the signs of tremors in the parental dams of the 30 mg/kg group). **Developmental NOEL:** 30 mg/kg/day (based upon the lack of treatment-related effects on the offspring of the 30 mg/kg group). **Study supplemental.** (Moore, 8/10/06)

TERATOLOGY, RABBIT

** 0061; 225370; "Study for Effects on Embryo-Fetal Development of S1264 Administered Orally to Rabbits"; (N. Horie; Environmental Health Science Laboratory, Sumitomo Chemical Co., Ltd., Konohana-Ku, Osaka, Japan; Study No. 3644; 7/26/02); Twenty-four artificially inseminated Kbl:NZW SPF female rabbits/group were dosed orally by gavage with 0 (vehicle: corn oil), 25, 125 or 250 mg/kg/day of S-1264 (lot no. PK-010501 G, purity: 94.9%) from day 6 through day 28 of gestation. One female each died in the 25, 125 and 250 mg/kg treatment groups on days 20, 23, and 14, respectively. The death of the female in the 25 mg/kg group was definitively associated with a dosing error, The female in the 125 mg/kg group was not pregnant. The does in the 250 mg/kg group demonstrated lower body weight gains and food consumption than did the control animals (NS). There was no treatment-related effect upon the development of the fetuses. **No adverse effect indicated. Maternal NOEL:** 125 mg/kg/day (based upon lower mean body weight gains and food consumption (not statistically significant) for the 250 mg/kg does); **Developmental NOEL:** 250 mg/kg/day (based upon the lack of a treatment-related effect at the 250 mg/kg treatment level); **Study acceptable.** (Moore, 8/11/06)

GENE MUTATION

** 0074; 225383; "Reverse Mutation Test of S-1264 in Bacterial Systems"; (S. Kitamoto; Environmental Health Science Laboratory, Sumitomo Chemical Co., Ltd, Osaka, Japan; Study No. 3673; 1/9/02); *S. typhimurium* strains TA98, TA100, TA1535 and TA1537 and *E. coli* strain WP2uvrA were treated with S-1264 (lot no. PK-010501G; purity: 94.9%) at concentrations ranging from 0 to 5000 ug/plate with a preincubation of 20 minutes and an incubation with plate incorporation for 48 hours at 37^{O} C under conditions of activation and non-activation. One trial was performed with triplicate samples for each treatment level. A phenobarbital and 5,6-benzoflavone-induced rat liver S9 fraction was used to metabolize the test material. There was no treatment-related increase in the incidence of reverse mutation. **No adverse effect indicated.** The positive controls were functional. **Study acceptable.** (Moore, 9/6/06)

CHROMOSOME EFFECTS

** 0077; 225386; "*In Vitro* Chromosomal Aberration Test on S-1264 in Chinese Hamster Lung Cells (CHL/IU)"; (K. Odawara; Environmental Health Science Laboratory, Sumitomo Chemical Co., Ltd, Osaka, Japan; Study No. 3633; 2/20/02); Chinese Hamster Lung cells (CHL/IU) were incubated with S-1264 (lot no. PK-010501G; purity: 94.9%) at concentrations ranging from 50 to 130 ug/ml (experiment 1, nonactivated), 20 to 110 ug/ml (experiment 2, non-activated), 50 to 250 ug/ml (experiment 1, activated) or 100 to 250 ug/ml (experiment 2, activated) at 37^{o} C. The non-activated samples were treated for 6 hours (experiment 1) with an additional 18 hours of incubation or for 24 hours (experiment 2). The activated samples received 6 hours of treatment and an additional 18 hours of incubation in both experiments. In both assays, the cells were incubated the last 1.5 hours with Colcemid prior to fixation. All of the incubations were performed with duplicate cultures. A phenobarbital and 5,6-benzoflavone-induced rat liver S9 fraction was used to metabolize the test material. There was no treatment-related increase in the percentage of cells with chromosomal aberrations under either assay condition. **No adverse effect indicated.** The positive controls were functional. **Study acceptable.** (Moore, 9/7/06)

DNA DAMAGE

** 0076; 225385; “Micronucleus Test on S-1264 in Mice”; (K. Odawara; Environmental Health Science Laboratory, Sumitomo Chemical Co., Ltd, Osaka, Japan; Study No. 3685; 7/26/02); Five male CD1 mice/group/time point were treated orally by gavage with 0 (corn oil) or 50.0 mg/kg of S-1264 (lot no. PK-010501G; purity: 94.9%) and euthanized at 24 and 48 hours post-dose. An additional 5 animals/group were dosed with 12.5 or 25.0 mg/kg and euthanized at 24 hours post-dose. As a positive control, another group of 5 male mice was treated by oral gavage with 60 mg/kg of cyclophosphamide and euthanized at 24 hours post-dose. The number of micronucleated polychromatic erythrocyctes (PCE) in 1000 PCEs and the ratio of PCEs to the total number of erythrocytes were reported. There was no treatment-related increase in the percentage of micronucleated PCEs. **No adverse effect indicated.** Positive control was functional. **Study acceptable.** (Moore, 9/7/06)

NEUROTOXICITY

Rat Acute Neurotoxity

53008-0066; 225375; “Oral (Gavage) Acute Neurotoxicity Study of S-1264 in Rats”; (R. G. York;
Argus Research, Horsham, PA and Pathology Associates Division, Charles River Laboratories, Inc., Discovery and Development Services, Frederick, MD; Project ID. 1119-032; 3/10/04); Ten Crl:CD (SD)IGS BR VAF/Plus rats/sex/group were dosed orally by gavage with 0 (vehicle: corn oil), 20, 50 or 100 mg/kg of S-1264 (lot no. PK-020301G, purity: 96.6%). Two males and 5 females in the 100 mg/kg group died or were euthanized in moribund condition within 24 hours of dosing. Clinical signs included whole body tremors and/or twitches and tachypnea/hyperpnea in the 100 mg/kg animals. These signs were observed shortly after dosing. Motor activity was increased in the 100 mg/kg animals at two hours post-dose. No treatment-related lesions were noted in the histological examination of the nervous tissues. **Possible adverse effect:** tremors and death; **Acute Neurotoxicity NOEL:** (M/F) 50 mg/kg (based upon the incidence of clinical signs noted in both sexes of the 100 mg/kg group). **Study acceptable.** (Moore, 8/28/06)

Rat Subchronic Neurotoxicity Study

53008-0067; 225376; “Oral (Diet) Subchronic Neurotoxicity Study of S-1264 in Rats”; (R. G. York; Argus Research, Horsham, PA and Pathology Associates Division, Charles River DDS - Worcester, Worchester, MA, Pathology Associates, a division of Charles River Laboratories, Inc., Frederick, MD, and Research Pathology Services, Inc., New Britain, PA; Project ID. 1119-033; 3/16/04); Twelve Crl:CD (SD)IGS BR VAF/Plus rats/sex/group received 0, 300, 1000 or 3000 ppm of S-1264 (lot no. PK-020301G, purity: 96.6%) in the diet for 13 weeks ((M) 0, 18.3, 59.8, 178.8 mg/kg/day, (F) 0, 20.9, 68.8, 206.0 mg/kg/day). One female in the 3000 ppm group died during the second week of treatment. The mean body weights and food consumption of both sexes in the 3000 ppm treatment group were lower than the control values over the course of the study ($p<0.01$ or 0.05). The incidence of soft and liquid feces was noted for the males in the 3000 ppm group ($p<0.01$). For the females in the 3000 ppm group, twitches and/or tremors were noted during the first two weeks of treatment ($p<0.01$). The FOB and motor activity assessments did not reveal any treatment-related effects. No treatment-related lesions were noted in the histopathological evaluation. **Possible adverse effect:** tremors. **Subchronic Neurotoxic NOEL:** (M/F) 1000 ppm ((M) 59.8 mg/kg/day, (F) 68.8 mg/kg/day) (based upon the incidence of possible neurologically-related signs noted for both sexes in the 3000 ppm treatment group). **Study acceptable.** (Moore, 8/30/06)

STUDIES ON METABOLITES

Gene Mutation

** 0075; 225384; “Reverse Mutation Test of MFFO in Bacterial Systems”; (S. Kitamoto; Environmental Health Science Laboratory, Sumitomo Chemical Co., Ltd, Osaka, Japan; Project ID. 3907; 9/30/04); *S. typhimurium* strains TA98, TA100, TA1535 and TA1537 and *E. coli* strain WP2uvrA were treated with S-1264 (lot no. PK-010501G; purity: 94.9%) at concentrations ranging from 4.88 to 156 ug/plate (non-activation, strains TA 100, TA 1535, TA 1537), from 19.5 to 625 ug/plate (activation, strains TA 100, TA 1535, TA 1537) and 156 to 5000 ug/plate (non-activation and activation, WP2uvrA, TA 98) with a preincubation of 20 minutes and an incubation with plate

incorporation for 48 hours at 37^{o} C under conditions of activation and non-activation. Two trials were performed with triplicate samples for each treatment level. A phenobarbital and 5,6-benzoflavone-induced rat liver S9 fraction was used to metabolize the test material. There was no treatment-related increase in the incidence of reverse mutation. **No adverse effect indicated.** The positive controls were functional. **Study acceptable.** (Moore, 9/6/06)

SUBCHRONIC STUDIES

Rat 4-Week Dietary Toxicity Study

53008-0054; 225363; "One-Month Oral Toxicity Study of S1264 in Rats"; (T. Kunimatsu; Environmental Health Science Laboratory, Sumitomo Chemical Co., Ltd., Konohana-Ku, Osaka, Japan; Study No. 3641; 6/27/02); Twelve Crj:CD (SD) rats/sex/group received 0, 300, 1000 or 3000 ppm of S-1264 (lot no. PK-010501 G, purity: 94.9%) in the diet for one month. Six animals/sex/group were dosed with 0 or 3000 ppm of the test material for the same time period and then maintained for an additional two weeks in a recovery phase ((M) 0, 28.6, 95.9, 284.5 mg/kg/day, (F) 0, 29.0, 95.2, 272.7 mg/kg/day). One female in the 3000 ppm group was found dead on day 4. The mean body weights of the males in the 1000 and 3000 ppm treatment groups were less than that of the control animals on day 8 of the study ($p<0.05$). No effect was evident thereafter. Mean food consumption for both sexes in the 3000 ppm group was less than those of the controls during the first week ($p<0.05$). No treatment-related effect upon water consumption was noted. The ophthalmology, urinalysis, and hematology examinations did not reveal any treatment-related effects. In the clinical chemistry, the total protein, albumin, and two of the globulins were elevated in the serum of the 3000 ppm males ($p<0.01$ or 0.05). This elevation of protein was not evident in the recovery animals. The total cholesterol and phospholipid levels were increased in the serum of the 1000 ppm males and both sexes in the 3000 ppm group ($p<0.01$). These effects also were not apparent at the end of the recovery period. The total bilirubin level was increased for the 3000 ppm males in both the treatment and recovery phases ($p<0.05$). The γ-GTP activity level was increased for both sexes in the 3000 ppm group ($p<0.01$). This effect was not noted in the recovery group. In the necropsy examination, the mean absolute and relative liver weights of both sexes in the 3000 ppm group were greater than those of the control ($p<0.01$). The mean relative liver weights of both sexes in the 1000 ppm group were also greater than those of the control ($p<0.01$ or 0.05). The increase in the mean relative liver weights of both sexes in the 3000 ppm group were still greater than those of the controls after 2 weeks of the recovery phase ($p<0.01$ or 0.05). The mean absolute and relative ventral prostate weights of the 3000 ppm males were less than those of the controls ($p<0.01$). No effect was noted in the recovery animals. The mean absolute and relative spleen weights of the 3000 ppm females were greater than those of the controls ($p<0.05$). No effect was evident in the recovery group. The mean relative thymus weight was greater than that of the control ($p<0.05$). This effect disappeared during the recovery phase. The histopathology examination revealed the presence of a diffuse hepatocellular hypertrophy in the liver of the 1000 ppm males and both sexes in the 3000 ppm group ((M) 0: 0/12 vs. 1000: 3/12, 3000: 10/12 ($p<0.01$), (F) 0: 0/12 vs. 3000: 7/12 ($p<0.01$). No lesion was evident in the livers of the recovery animals. In the liver tissue which was examined by electron microscopy, smooth endoplasmic reticulum (SER) was increased in the livers of the two 3000 ppm males in the treatment group. This increase in SER was not evident for the 3000 ppm females in the treatment group or either sex in the recovery group. Target organ: liver; **No adverse effect indicated. One Month Dietary NOEL:** (M/F) 300 ppm ((M) 28.6 mg/kg/day, (F) 29.0 mg/kg/day) (based uponthe increased mean relative liver weights of both sexes in the 1000 ppm treatment group and other effects on the liver noted for the males in this group). **Study supplemental.** (Moore, 8/1/06)

Rat Subchronic Dietary Toxicity Study

53008-0051; 225360; "S-1264: 13-Week Repeated Dose Oral Toxicity (Feeding) Study in the Wistar Rat"; (E.W. Sommer, C. Knuppe, P. Gretener, K. Weber; RCC Ltd., Toxicology, 4332 Stein, Switzerland; Study No. 841950; 9/19/03); Twelve Wistar rats/sex/group received 0, 100, 300, 1000 or 2500 ppm of S-1264 (lot no. PK-010501 G, purity: 94.9%) in the diet for 13 weeks

((M) 0, 6.85, 20.6, 70.4 and 183.6 mg/kg/day, (F) 0, 7.51, 21.6, 73.0, 185.6 mg/kg/day). One female in the 2500 ppm group died on day 4. The mean body weights of both sexes in the 2500 ppm group were less than those of the control group ((M) $p<0.01$, (F) NS). The Functional Observational Battery did not reveal any treatment-related signs. There was no treatment-related effect upon food consumption. The ophthalmology examination did not reveal any treatment-related effect. Although the number and relative concentration of basophils in the blood of the 2500 ppm females was less than that of the controls, no treatment-related effect was apparent in the overall hematology profile for these animals. The mean serum cholesterol and phospholipid levels were elevated for the males in the 1000 ppm group and for both sexes in the 2500 ppm group ($p<0.01$). The total protein and albumin levels in the serum were increased for the 2500 ppm males ($p<0.01$). The 1000 ppm males also demonstrated an increase in total protein ($p<0.01$). The males in the 1000 and 2500 ppm groups exhibited increased levels of leucocytes in the urine ($p<0.01$). The mean absolute and relative liver weights of the 1000 ppm males and both sexes in the 2500 ppm group were greater than those values for the control group ($p<0.01$). The mean relative kidney weights were greater for both sexes in the 2500 ppm group ($p<0.01$ or 0.05). The mean relative spleen weight of the 2500 ppm males was greater than that of the control ($p<0.05$). In the histopathology examination, hepatocellular hypertrophy and basophilia in the liver were evident for both the sexes in the 2500 ppm group and for the males in the 300 and 1000 ppm groups (hepatocellular hypertrophy: (M) 0: 1/12 vs. 300: 5/12, 1000: 10/12, 2500: 12/12, (F) 0: 0/12 vs. 2500: 5/12; hepatocellular basophilia: (M) 0: 0/12 vs. 300: 5/12, 1000: 12/12, 2500: 12/12; (F) 0: 0/12 vs. 2500: 4/12). An increasing severity of hyaline droplet formation was noted in the kidneys of the 2500 ppm males (grade 3: 0: 1/12 vs. 2500: 7/12). Target organs: liver and kidneys; **No adverse effect evident. Subchronic dietary NOEL:** (M 100 ppm (6.9 mg/kg/day) (based upon the incidence of hepatocellular hypertrophy and basophilia in the livers of the 300 ppm males), (F) 300 ppm (21.6 mg/kg/day) (based upon increased relative liver weights for the females in the 1000 ppm group); **Study acceptable.** (Moore, 7/25/06)

Rat 6-Month Dietary Toxicity Study

53008-0055; 225364; "Six-Month Oral Toxicity Study of S1264 in Rats"; (T. Kunimatsu; Environmental Health Science Laboratory, Sumitomo Chemical Co., Ltd., Konohana-Ku, Osaka, Japan; Study No. 3663; 7/31/02); Twelve Crj:CD (SD) rats/sex/group received 0, 300, 1000 or 3000 ppm of S-1264 (lot no. PK-010501 G, purity: 94.9%) in the diet for 6 months ((M) 0, 5.3, 16.0, 54.1, 164.6 mg/kg/day, (F) 0, 6.4, 19.0, 65.4, 191.4 mg/kg/day). No deaths occurred during the study. During the first week of treatment, all of the animals in the 3000 ppm group demonstrated tremors. Thereafter, tremors were only observed in two males and one female of this group during the 2nd and 3rd weeks, respectively. The mean body weights of the treated animals were not affected by the treatment. There was no treatment-related effect on food consumption except during the first week of treatment when the mean food consumption of the 3000 ppm males was less than that of the control ($p<0.01$). The ophthalmology, urinalysis and hematology evaluations did not reveal any treatment-related effects. In the clinical chemistry, the total serum protein and albumin were elevated for the 1000 and 3000 ppm males ($p<0.01$ or 0.05). The β-G1 b globulin of the 3000 ppm males was increased as well ($p<0.05$). The total cholesterol and phospholipid levels of the 1000 and 3000 ppm males were greater than the control values ($p<0.01$). The total bilirubin in the serum of the 3000 ppm males was greater than that of the control ($p<0.01$). In contrast, the total bilirubin level in the 1000 and 3000 ppm females was less than that of the controls ($p<0.05$). The γ-GTP activity in the serum of both sexes in the 3000 ppm group was greater than that of the control ($p<0.01$ or 0.05). The mean relative liver weights of the 1000 ppm males and both sexes of the 3000 ppm group and the mean absolute liver weight of the 3000 ppm males were greater than the control values ($p<0.01$). The histopathology examination revealed the presence of a diffuse hepatocellular hypertrophy in the livers of both sexes in the 1000 ppm and 3000 ppm groups ((M) 0: 0/12 vs. 1000: 9/12, 3000: 12/12 ($p<0.01$), (F) 0: 0/12 vs. 1000: 4/12, 3000: 10/12 ($p<0.01$). An increased incidence of microvesicular steatosis was noted in the livers of the 3000 ppm males (0: 2/12 vs. 3000: 9/12 ($p<0.01$)). In the liver tissue which was examined by electron microscopy, smooth endoplasmic reticulum (SER) was increased in the livers of the two 3000 ppm males in the treatment group. This increase in SER was not evident for the 3000 ppm females. Target organ: liver; **Possible adverse effect:** tremors. **Subchronic Dietary Toxicity NOEL:** (M/F) 300 ppm ((M) 16.0

mg/kg/day, (F) 19.0 mg/kg/day) (based upon the lesions in the livers of both sexes in the 1000 ppm group and the other liver-related effects noted for the 1000 ppm males). **Study acceptable.** (Moore, 8/2/06)

Mouse Subchronic Dietary Toxicity Study

53008-0052; 225361; "S-1264: 13-Week Repeated Dose Oral Toxicity (Feeding) Study in the CD-1 Mouse"; (E.W. Sommer, C. Knuppe, P. Gretener, K. Weber; RCC Ltd., Toxicology, CH-4452 Itingen, Switzerland; Study No. 841949; 2/23/04); Twelve CD-1 mice/sex/group received 0, 100, 1500, 2500 or 3500 ppm of S-1264 (lot no. PK-010501 G, purity: 94.9%) in the diet for 13 weeks ((M) 0, 13.7, 208.8, 357.0 and 486.9 mg/kg/day, (F) 0, 17.2, 252.1, 439.1, 587.4 mg/kg/day). One male in the control group was euthanized on day 76. One female each in the 1500, 2500 and 3500 ppm groups, respectively, died on day 92, shortly after blood collection. There was no treatment-related effect upon mean body weights or food consumption. The hematology examination did not reveal any treatment-related effect. The mean serum cholesterol levels were elevated for the females in the 2500 and 3500 ppm groups ($p<0.01$, 0.05). The mean serum phospholipid level for the 3500 ppm females was elevated as well ($p<0.01$). The albumin levels in the serum were increased for the 2500 and 3500 ppm males ($p<0.01$). The mean absolute liver weights of the males in the 1500 and 2500 ppm groups and both sexes in the 3500 ppm group were greater than those values for the control group ($p<0.01$). The mean relative liver weights of the 1500 ppm males and of both sexes in the 2500 and 3500 ppm groups were greater than the control values ($p<0.01$, 0.05). In the histopathology examination, hepatocellular hypertrophy in the liver was evident for both the sexes in the 1500, 2500 and 3500 ppm groups ((M) 0: 1/12 vs. 1500: 4/12, 2500: 7/12, 3500: 7/12, (F) 0: 0/12 vs. 1500: 8/12, 2500: 11/12, 3500: 9/12). Hepatic necrosis was noted for both sexes in the 3500 ppm group ((M/F) 0: 0/12 vs. 3500: 3/12). Target organ: liver; **Possible adverse effect**: hepatic necrosis. **Subchronic Dietary Toxicity NOEL:** (M/F) 100 ppm ((M) 13.7 mg/kg/day, (F) 17.2 mg/kg/day) (based on the hepatocellular hypertrophy noted for both sexes in the 1500 ppm treatment group); **Study supplemental** (current guideline requirements for a subchronic toxicity study were not fulfilled). (Moore, 7/26/06)

Dog Subchronic Oral Toxicity Study

53008-0053; 225362; "90-Day Oral Toxicity Study with S-1264 in Beagle Dogs Followed by 42-Day Recovery Study"; (H. Uchida; Panapharm Laboratories Co., Ltd., Uto-shi, Kumamoto, 869-0425, Japan; Study No. 201142; 7/30/02); Four beagle dogs/sex/group were dosed orally by capsule with 0, 10, 30 or 100 mg/kg/day of S-1264 (lot no. PK-010501 G; purity: 94.9%) for 13 weeks. An additional two animals/sex/group in the control and the 100 mg/kg groups were treated for the 13 weeks and then maintained as recovery animals for 6 weeks post-treatment. Both sexes in the 30 and 100 mg/kg groups demonstrated an increased incidence of emesis over the course of the treatment. In addition, both sexes in the 100 mg/kg group sporadically suffered tremors during the treatment. No effects were evident during the recovery period. There was no apparent treatment-related effect upon the mean body weights or food consumption. The ophthalmology examination did not reveal any treatment-related effect. No treatment-related effects were evident in the hematology, clinical chemistry or urinalysis. The mean absolute or relative organ weights were not affected by the treatment. No treatment-related lesions were noted in the histopathological examination. **Possible adverse effect:** tremors. **Subchronic Oral Toxicity NOEL:** (M/F) 10 mg/kg/day (based upon the incidence of treatment-related signs in the 30 mg/kg treatment group); **Study acceptable.** (Moore, 7/28/06)

Rat Repeated Dosing Subchronic Dermal Toxicity Study

53008-0056; 225365; "A 90-Day Repeated Dose Dermal Toxicity Study of S1264 in Rats"; (H. Furukawa; Safety Assessment Laboratory, Panapharm Laboratories Co., Ltd., Uto-shi, Kumamoto 869-0425, Japan; Study No. P030373; 6/24/04); The skin of twelve Crj:CD (SD) rats/sex/group was exposed to 0, 30, 100, 300 or 1000 mg/kg/day of S-1264 (lot no. PK-020301 G, purity: 96.6%), 6 hours/day, for 90 days under an occlusive wrap. Two females in the 1000 mg/kg group died on day 4. One male in the same group was euthanized *in extremis* on day 73. Vocalization and hyperactivity were noted as clinical signs 30 minutes and 3 to 5 hours post-application for 4 females in the 30 mg/kg group, 10 females and one male in the 100 mg/kg group and 10 females and 3 males each in the 300 and 1000 mg/kg groups on the first day of treatment. These signs

persisted in a decreasing number of animals through day 4. Thereafter, the signs were not observed. There was no treatment-related effect upon the body weights or food consumption. The ophthalmology and hematology examinations did not reveal any treatment-related effects. In the clinical chemistry, the males in the 1000 mg/kg group demonstrated an increased level of activity for γ-GTP in the serum ($p<0.05$). The mean relative liver weight of the 1000 mg/kg males was greater than that of the controls ($p<0.01$). The females in the 1000 mg/kg group demonstrated the presence of squamous cell hyperplasia on the skin which had been treated (0: 0/12 vs. 1000: 5/10, $p<0.01$). **No adverse effect indicated. Subchronic Dermal Systemic NOEL:** (M) 30 mg/kg/day (based upon the presence of treatment-related signs in the 100 mg/kg group (vocalization and hyperactivity), (F) < 30 mg/kg/day (based upon the presence of treatment-related signs in the 30 mg/kg group (vocalization and hyperactivity); **Localized Dermal Irritation NOEL:** (M) 1000 mg/kg/day (based on the lack of treatment-related effects on the treated skin), (F) 300 mg/kg/day (based upon the incidence of squamous cell hyperplasia on the treated skin of the 1000 mg/kg group); **Study acceptable.** (Moore, 8/3/06)

Rat 4-Week Inhalation Toxicity Study

53008-0057; 225366; "Four-Week Repeated Inhalation Toxicity Study of S1264 in Rats"; (Y. Deguchi; Environmental Health Science Laboratory, Sumitomo Chemical Co., Ltd., Konohana-Ku, Osaka, Japan; Study No. 3704; 8/26/02); Ten Crj:CD (SD) rats/sex/group were exposed nose-only to 0 (corn oil vehicle), 9.84, 50.6, 98.7 and 196 mg/m^3 (analytical) of S-1264; lot no. PK-010501 G, purity: 94.9%) 4 hours/day for 4 weeks. The mean MMAD (GSD) values were 2.67 (2.17), 3.02 (2.00), 2.47 (2.02) and 2.94 (2.25) um, respectively. Seven males and three females in the 196 mg/m^3 died during the study. Clinical signs of tremors, hypersensitivity and clonic convulsions were noted for the animals in this group, particularly during the first week of the exposure. Six of the 10 animals which died succumbed during the first week. There was no treatment-related effect upon the mean body weights. The mean food and water consumption for the both sexes in the 196 mg/m^3 exposure group were less than those values for the control group ($p<0.01$, 0.05 or NS) during the first week of exposure. There was no effect on these parameters during the remainder of the exposure period. The ophthalmology, hematology, clinical chemistry and urinalysis did not reveal any treatment-related effects. The necropsy and histopathological examinations did not reveal any treatment-related lesions or effect on organ weights. **Possible adverse effect:** tremors and clonic convulsions; **4-Week Inhalation Exposure NOEL:** (M/F) 98.7 mg/m^3 (based upon the clinical signs demonstrated by both sexes in the 196 mg/m^3 exposure group); **Study supplemental.** (Moore, 8/4/06)

MECHANISTIC STUDIES

53008-0069; 225378; "Study of the Mode of Action of S-1264 for Liver Tumor Promotion in Rats"; (Y. Deguchi; Environmental Health Science Laboratory, Sumitomo Chemical Co., Ltd., Osaka, Japan; Study No. S1226; 6/13/05); At least 5 BrlHan:WIST rats/sex/group received 0, 200, 900, 1800, or 3600 ppm of S-1264 (lot no. PK-020301 G, purity: 96.6%) in the diet for 7 days. An additional 5 animals/sex/group were treated with 0, 1800 or 3600 ppm of the test material in the same manner and then kept on a maintenance diet for 7 days (Treated Cohort: (M) 0, 12.7, 63.5, 121.1, 220.8, mg/kg/day, (F) 0, 13.2, 57.6, 95.7, 149.3 mg/kg/day; Recovery Cohort: M) 0, 123.3, 255.7 mg/kg/day, (F) 0, 145.7, 280.9 mg/kg/day). An additional 10 animals/sex were treated with 1000 ppm of phenobarbital (PB) as a positive control group in the same manner with five animals being retained as part of the recovery group (Treated Cohort: (M) 63.9 mg/kg/day, (F) 66.9 mg/kg/day; Recovery Cohort: (M) 67.3 mg/kg/day, (F) 78.0 mg/kg/day). Two females and one male in the 3600 ppm group died during the treatment period. In the Treated Cohort, the mean absolute liver weights of both sexes treated with 1000 ppm PB and the mean relative liver weights of the females treated with 1800 and 3600 ppm and both sexes treated with PB were increased over the control values ($p<0.01$ or 0.05). Light microscopic examination of the liver tissue derived from animals in the Treated Cohort revealed slight centrilobular hepatocytic hypertrophy in the livers of both sexes in the 1800 and 3600 ppm groups and mild to moderate hypertrophy in the PB treated animals ((M) 0: 0/5 vs. 1800: 1/5, 3600: 2/7, PB, 1000: 4/5; (F) 0: 0/5 vs. 1800: 1/5, 3600: 2/5, PB, 1000: 3/5). In the Recovery Cohort, one male in the 3600 ppm group and two of the PB animals/sex retained a slight centrilobular hypertrophy 7 days after

cessation of treatment. In the electron microscopic examination of liver tissue from two animals/sex in the control, 3600 ppm group and the 1000 ppm PB group of both the treated and recovery cohorts, slight to mild dilatation/proliferation of the smooth endoplasmic reticulum was noted for the animals in the 3600 ppm treatment group and mild to moderate for the animals in the 1000 ppm PB group. In the BrdU labeling study, the males in the 900 and 1800 ppm groups and both sexes in the PB group of the Treated Cohort demonstrated an increased percentage of labeled hepatocytes. Seven days after cessation of treatment, the labeling had declined to less than that of the control animals. Analysis of the liver mRNA and protein in the Treated Cohort of the PB animals revealed an increase in the total microsomal protein for the males and increases in the total P450 protein and the relative mRNA levels of CYP2B1/2, CYP3A1 and CYP3A2 and protein level for CYP2B for both sexes ($p<0.01$ or 0.05). The mRNA levels of CYP2B1/2 and CYP3A1 and the protein level for CYP2B were increased for both sexes in the 3600 ppm Treated Cohort. The mRNA levels of CYP2B1/2 were increased for both sexes in the 1800 ppm group and for the females in the 900 ppm group of the Treated Cohort. These effects were not evident 7 days after dosing. **Study supplemental.** (Moore, 8/31/06)

53008-0070; 225379; "The 2nd Study for Mode of Action of S-1264 for Liver Tumor Promotion in Rats'; (Y. Deguchi; Environmental Health Science Laboratory, Sumitomo Chemical Co., Limited, Osaka, Japan; Study No. S1255; 1/24/06); Eight BrlHan:WIST rats/sex/group received 0, 200, 900, 1800, or 3600 ppm of S-1264 (lot no. PK-020301 G, purity: 96.6%) in the diet for 7 days. An additional 8 animals/sex/group were treated with 0, 1800 or 3600 ppm of the test material in the same manner and then kept on a maintenance diet for 7 days (Treated Cohort: (M) 0, 13.5, 61.9, 126.9, 253.8 mg/kg/day, (F) 0, 14.5, 64.5, 143.7, 262.5 mg/kg/day; Recovery Cohort: M) 0, 126.5, 245.4 mg/kg/day, (F) 0, 130.4, 246.6 mg/kg/day). An additional 16 animals/sex were treated with 1000 ppm of phenobarbital (PB) as a positive control group in the same manner with 8 animals being retained as part of the recovery group (Treated Cohort: (M) 67.7 mg/kg/day, (F) 74.3 mg/kg/day; Recovery Cohort: (M) 67.0 mg/kg/day, (F) 75.2 mg/kg/day). Three females in the 3600 ppm group died during the treatment period. Tremors were noted for a few animals in the 3600 ppm group during the treatment period. In the Treated cohort, the mean absolute liver weights for the 1800 ppm males, the 3600 ppm females and for both sexes in the 1000 ppm PB group were greater than the control values ($p<0.01$ or 0.05). The mean relative liver weights for the 1800 ppm males and for both sexes in the 3600 ppm and 1000 ppm PB groups were greater than the control values ($p<0.01$). In the Recovery Cohort, the mean absolute liver weights for the 1000 ppm PB males and the mean relative liver weights for the 1800 ppm males and the 1000 ppm PB males were greater than the control values ($p<0.05$). In the histopathological evaluation, centrilobular hypertrophy of the hepatocytes in the liver was evident for the both sexes in the 1000 ppm PB group in both the Treated and Recovery Cohorts ((M) Treated, 0: 0/8 vs. 1000: 8/8 ($p<0.01$), Recovery, 0: 0/8 vs. 1000: 4/8 ($p<0.05$); (F) Treated, 0: 0/8 vs. 1000: 8/8 ($p<0.01$), Recovery, 0: 0/8 vs. 1000: 1/8 (NS)). For the animals dosed with the test material, hepatocytic hypertrophy was noted for one male in the 1800 ppm group and two males and one female in the 3600 ppm group of the Treated Cohort. These effects were not evident for the Recovery Cohort. Decreased vacuolation was noted in the liver hepatocytes of the 1800 ppm males and both sexes of the 3600 ppm group in the Treated Cohort ((M) 0: 0/8 vs. 1800: 1/8, 3600: 5/8 ($p<0.01$), (F) 0: 0/8 vs. 3600: 2/8 (NS)). This effect was not noted for the 1000 ppm PB group. In the hepatic gap junction intercellular communication (GJIC) assay, *ex vivo* dye transfer was reduced for both sexes in the 1800 and 3600 ppm groups and the 1000 ppm PB group of the Treated Cohort ($p<0.01$ or NS). The effect was not evident in the Recovery Cohort. In other parameters which were evaluated, lipid peroxidation was not affected for the animals treated with the test material. The females in the 1000 ppm PB Treated Cohort had a lower level of lipid peroxidation ($p<0.01$). The males in the positive control group of the Recovery Cohort had a higher lipid peroxidation level ($p<0.01$). The total GSH levels for both sexes of the 900, 1800 and 3600 ppm groups in the Treated Cohort were increased over the control values ($p<0.01$ or 0.05). The reduced GSH levels for the 900 ppm males and for both sexes in the 1800 and 3600 ppm groups in the Treated Cohort were increased as well ($p<0.01$). The reduced GSH levels persisted in both sexes of the 3600 ppm group in the Recovery Cohort ($p<0.01$ or 0.05). The total GSH levels in the 1000 ppm PB males of the Treated and Recovery Cohorts were elevated as well ($p<0.05$). The total GSH levels for the 1000 ppm PB males in the Recovery Cohort and the 1000 ppm females in the

Treated Cohort were greater than the control values ($p<0.01$). Apoptosis was decreased only in the 1000 ppm males of the Treated Cohort ($p<0.01$). An overall comparison of the study results indicates that the test material had some effects which were similar to those observed for phenobarbital (i.e., hepatic hypertrophy, decreased GJIC). A greater degree of oxidative stress was noted for the animals treated with the test material (greater total and reduced GSH levels). Alteration of apoptosis was not apparently affected by the treatment. **Study supplemental.** (Moore, 9/5/06)

53008-0071; 225380; "Study for Mode of Action of S-1264 for Liver Tumor Promotion in Rats (*In Vitro* Effect of S-1264 on Cytochrome P450 Activity and mRNA Level)"; (H. Nagahori; Environmental Health Science Laboratory, Sumitomo Chemical Co., Limited, Osaka, Japan; Study No. X0145; 1/31/06). Primary hepatocyte cultures of male BRLHan:WIST rats and male crlj:CD 1 (ICR) mice and fresh female human hepatocytes were exposed to 50 uM of S-1264 (purity not reported) or 50 uM of phenobarbital for 3 days. The activity of 7-pentoxyresorufin O-dependentylase and the CYP2B mRNA content in the hepatocytes were assayed at the conclusion of the incubation period. Treatment with S-1264 resulted in an increase in 7-pentoxyresorufin O-dependentylase activity and a 2 to 3 fold increase in the CYP2B mRNA content above that of the control levels for the rat and human hepatocyte cultures. The test material did not demonstrate any effect upon the mouse hepatocytes. Phenobarbital increased the enzyme activity and the mRNA levels in all of the hepatocyte cultures. **Supplemental Study.** (Moore, 9/5/06)

53008-0073; 225382; "Gene Expression Profiling Analysis of Early Phase of Treatment in the Liver from S-1264-Treated Rats"; (T. Yamada; Environmental Health Science Laboratory, Sumitomo Chemical Co., Limited, Osaka, Japan; Study No. S1274; 1/31/06); The gene expression profile of liver samples from male BrlHan:WIST rats which were fed 1800 ppm of S-1264 (purity not reported) or 1000 ppm of phenobarbital (PB) in the diet for one week were analyzed. The numbers of probe sets which demonstrated a 2-fold change were 25 and 85 for S-1264 and PB, respectively in the up-regulation mode and 10 and 14, respectively, in the down-regulation mode. Twenty-one of the 25 up-regulated by S-1264 were also common to the PB data set. Four of the 10 sets down-regulated by S-1264 were also down-regulated by PB. Among the genes up-regulated by both S-1264 and PB were glutathione-S-transferase, CYP2B, UDP glycosyltransferase, aflatoxin B1 aldehyde reductase, epoxide hydroxylase, and liver UDP-glucuronosyltransferase. **Study supplemental.** (Moore, 9/5/06)

METABOLISM STUDIES

Metabolism, Rat

** 53008-0078; 225387; "The Disposition and Metabolism of [Carbonyl-^{14}C] S-1264RTZ in Rats"; (K. Sugimoto; Laboratory of Pharmacokinetics and Pharmacology, Panapharm Laboratories Co., Ltd., Kumamoto, 869-0425, Japan; Study No. PK0141; 7/31/02, amended, 4/9/04); Sprague-Dawley rats of both sexes were treated with 1 or 20 mg/kg of [Carbonyl-^{14}C] S-1264RTZ (lot no. RIS2000-019, specific radioactivity: 6.50 Mbq/mg; radiochemical purity: (12/00) 99.4%; repurified 4 times during study, (12/3/01) 99.6%, (12/19/1) 98.2%, (2/18/02) 98.6%, (2/25/02) 98.9%). Dosing preparations were supplemented with unlabeled S-1264RTZ (lot no. 010621G, purity: 99.3%). In the Excretion Study, four animals/sex/group were dosed orally by gavage with 1 or 20 mg/kg of the test material and urine and fecal samples were collected up to 7 days post-dose. Air samples were collected through 72 hours. In the Biliary Excretion Study, four male rats, whose bile ducts had been cannulated, were dosed with 1 mg/kg of the test material. Bile, urine and fecal samples were collected through 72 hours post-dose. In the Pharmacokinetics Study, 3 animals/sex/group were dosed with 1 or 20 mg/kg of the test material and blood samples were drawn at specified time intervals up to 7 days post-dose. In the Tissue Distribution Study, 12 animals/sex/group were dosed with 1 or 20 mg/kg of the test material and 3 animals/sex/group/time point were euthanized at specified time intervals post-dose and the concentration of radiolabel in particular tissues was determined. In the Excretion Study, 44 to 57% of the administered dose was excreted in the urine and 38 to 52% in the feces with little difference demonstrated between the two dosing levels. Air sampling recovered from 0.7 to 1.4% of the administered dose. Seventy four to 83% of the administered dose was recovered in the 1st 24 hours post-dose. In the Bile Excretion Study, 30 to 40% of the dose was recovered in the bile,

25 to 26% in the urine and 30 to 39% in the feces. The males and females demonstrated 58 and 68% absorption of the administered dose. The time to maximal blood concentrations ranged from 3.3 to 6.7 hours post-dose. The liver and kidneys were the primary sites of radiolabel recovery over the course of the 7 day collection period. Radioassay of the stomach, small intestine, cecum and large intestine demonstrated a time-course of passage for the radiolabeled compound through the gastrointestinal tract. Radiolabel recovery in the hair peaked later than that observed in the systemic circulation. The radiolabel was not sequestered in the fat nor did a significant fraction of the administered dose pass across the blood:brain barrier. Based upon the isolated metabolites, the major pathways of metabolism were cleavage of the ester linkage, oxidation of methyl groups in the acid moiety of the molecule to form alcohol, aldehyde and carboxylic acid metabolites, reduction of the double bond in the acid moiety of the molecule, and the formation of glutathione, sulfate and glucuronide adducts. **Study acceptable.** (Moore, 9/25/06)

** 53008-0079; 225388; "The Disposition and Metabolism of [Carbonyl-^{14}C] S-1264RTE in Rats"; (K. Sugimoto; Laboratory of Pharmacokinetics and Pharmacology, Panapharm Laboratories Co., Ltd., Kumamoto, 869-0425, Japan; Study No. PK0143; 7/31/02, amended, 4/9/04); Sprague-Dawley rats of both sexes were treated with 1 or 20 mg/kg of [Carbonyl-^{14}C] S-1264RTE (lot no. RIS2000-020, specific radioactivity: 6.50 Mbq/mg; radiochemical purity: (12/00) 99.2%; repurified 4 times during study, (3/18/02) 99.0%, (3/26/02) 98.1%, (4/15/02) 99.4%, (4/22/02) 99.2%;). Dosing preparations were supplemented with unlabeled S-1264RTE (lot no. 010831G, purity: 96.9%) . In the Excretion Study, four animals/sex/group were dosed orally by gavage with 1 or 20 mg/kg of the test material and urine and fecal samples were collected up to 7 days post-dose. Air samples were collected through 72 hours. In the Biliary Excretion Study, four male rats, whose bile ducts had been cannulated, were dosed with 1 mg/kg of the test material. Bile, urine and fecal samples were collected through 72 hours post-dose. In the Pharmacokinetics Study, 3 animals/sex/group were dosed with 1 or 20 mg/kg of the test material and blood samples were drawn at specified time intervals up to 7 days post-dose. In the Tissue Distribution Study, 12 animals/sex/group were dosed with 1 or 20 mg/kg of the test material and 3 animals/sex/group/time point were euthanized at specified time intervals post-dose and the concentration of radiolabel in particular tissues was determined. In the Excretion Study, 29 to 45% of the administered dose was excreted in the urine and 50 to 66% in the feces with little difference demonstrated between the two dosing levels. Air sampling recovered 0.4 to 0.6% of the administered dose. Sixty five to 78% of the administered dose was recovered in the 1st 24 hours post-dose. In the Bile Excretion Study, 27 and 55% of the dose was recovered in the bile of the males and females, respectively. The greater recovery of the radiolabel in the bile of the females resulted in a decrease of radioactivity recovered from the urine and the feces (urine: 45.1 to 20.4%, feces: 50.2 to 19.6%), For the males, only the urinary recovery of the radiolabel was decreased (37.1 to 9.6%). The males absorbed only 40% of the administered dose in contrast to the 77% demonstrated by the females. The time to maximal blood concentrations ranged from 4.7 to 6.7 hours post-dose, irrespective of the dosing level. The liver was the primary site of radiolabel recovery over the course of the 7 day collection period. Radioassay of the stomach, small intestine, cecum and large intestine demonstrated a time-course of passage for the radiolabeled compound through the gastrointestinal tract. Radiolabel recovery in the hair peaked later than that observed in the systemic circulation. The radiolabel was not sequestered in the fat nor did a significant fraction of the administered dose pass across the blood:brain barrier. Based upon the isolated metabolites, the major pathways of metabolism were cleavage of the ester linkage, oxidation of methyl groups in the acid moiety of the molecule to form alcohol, aldehyde and carboxylic acid metabolites, reduction of the double bond in the acid moiety of the molecule, and the formation of a sulfate adduct. **Study acceptable.** (Moore, 10/17/06)

53008-0080; 225389; "The Disposition and Metabolism of [methoxymethylbenzyl-α-^{14}C] S-1264RTZ in Rats"; (K. Sugimoto; Laboratory of Pharmacokinetics and Pharmacology, Panapharm Laboratories Co., Ltd., Kumamoto, 869-0425, Japan; Study No. PK0142; 7/31/02, amended, 4/9/04); Sprague-Dawley rats of both sexes were treated with 1 or 20 mg/kg of [Methoxymethylbenzyl-α -^{14}C] S-1264RTZ (lot no. RIS2001-003, specific radioactivity: 6.19 Mbq/mg; radiochemical purity: (12/00) 99.4%; repurified 4 times during study, (10/22/01) 99.1%, (10/31/01) 98.6%, (11/19/01) 99.1%, (11/26/01) 99.5%). Dosing preparations were supplemented

with unlabeled S-1264RTZ (lot no. 010621G, purity: 99.3%). In the Excretion Study, four animals/sex/group were dosed orally by gavage with 1 or 20 mg/kg of the test material and urine and fecal samples were collected up to 7 days post-dose. Air samples were collected through 72 hours. In the Pharmacokinetics Study, 3 animals/sex/group were dosed with 1 or 20 mg/kg of the test material and blood samples were drawn at specified time intervals up to 7 days post-dose. In the Tissue Distribution Study, 12 animals/sex/group were dosed with 1 or 20 mg/kg of the test material and 3 animals/sex/group/time point were euthanized at specified time intervals post-dose and the concentration of radiolabel in particular tissues was determined. In the Excretion Study, 60 to 71% of the administered dose was excreted in the urine and 25 to 36% in the feces with little difference demonstrated between the two dosing levels. No radiolabel was recovered in the air sampling. Sixty eight to 84% of the administered dose was recovered in the 1st 24 hours post-dose. The time to maximal blood concentrations ranged from 4.0 to 8.0 hours post-dose. The liver and kidneys were the primary sites of radiolabel recovery over the course of the 7 day collection period. Radioassay of the stomach, small intestine, cecum and large intestine demonstrated a time-course of passage for the radiolabeled compound through the gastrointestinal tract. Radiolabel recovery in the hair peaked later than that observed in the systemic circulation. The radiolabel was not sequestered in the fat nor did a significant fraction of the administered dose pass across the blood:brain barrier. Based upon the isolated metabolites, the major pathways of metabolism were cleavage of the ester linkage, oxidation of methyl groups in the acid moiety of the molecule to form alcohol, aldehyde and carboxylic acid metabolites, reduction of the double bond in the acid moiety of the molecule, and the formation of glutathione, sulfate and glucuronide adducts. **Study supplemental.** (Moore, 10/19/06)

53008-0081; 225390; "The Disposition and Metabolism of [Carbonyl-^{14}C] S-1264RTZ (1R-trans-Z) after Repeated Administration to Rats"; (K. Sugimoto; Laboratory of Pharmacokinetics and Pharmacology, Panapharm Laboratories Co., Ltd., Kumamoto, 869-0425, Japan; Study No. P020096; 9/11/02); Male Sprague-Dawley rats were treated with 1 mg/kg/day of [Carbonyl-^{14}C] S-1264RTZ (lot no. RIS2000-019, specific radioactivity: 6.50 Mbq/mg; radiochemical purity: (12/00) 99.4%; repurified 2 times during study;(5/8/02) 98.6%, (5/16/02) 99.7%),. Dosing preparations were supplemented with unlabeled S-1264RTZ (lot no. 010621G, purity: 99.3%). In the Excretion Study, three animals were dosed orally by gavage daily for 21 days with the test material and urine and fecal samples were collected up daily through 21 days and at 24, 48, and 72 hours and 5, 7, 10 and 14 days post-final dose. In the Tissue Distribution Study, 12 animals were dosed orally by gavage with the test material and 3 animals/time point were euthanized after 10, 16, and 21 doses and 7 days post-final dose. In the Excretion Study, 57% of the total administered dose was excreted in the urine and 38% in the feces. The liver and kidneys were the primary sites of radiolabel recovery over the course of the dosing period with the level of radioactivity stabilizing over the dosing period. However, at 14 days post-final dose, the red blood cell and hair were the primary locations where the radiolabel was recovered. The level of radioactivity recovered from the stomach, small intestine, cecum and large intestine was stabilized over the course of the dosing period. Radiolabel recovered in the hair peaked at the time of the 21st dose. The radiolabel was not sequestered in the fat nor did a significant fraction of the administered dose pass across the blood:brain barrier. Based upon the isolated metabolites, the major pathways of metabolism were cleavage of the ester linkage, oxidation of methyl groups in the acid moiety of the molecule to form alcohol, aldehyde and carboxylic acid metabolites, reduction of the double bond in the acid moiety of the molecule, and the formation of glutathione, sulfate and glucuronide adducts. **Study supplemental.** (Moore, 10/20/06)

53008-0082; 225391; "The Disposition and Metabolism of [Methoxymethylbenzyl-α-^{14}C] S-1264RTZ (1R-trans-Z) after Repeated Administrations to Rats"; (K. Sugimoto; Laboratory of Pharmacokinetics and Pharmacology, Panapharm Laboratories Co., Ltd., Kumamoto, 869-0425, Japan; Study No. P020095; 9/11/02, amended, 4/9/04); Male Sprague-Dawley rats were treated with 1 mg/kg/day of [Methoxymethylbenzyl-α -^{14}C] S-1264RTZ (lot no. RIS2001-003, specific radioactivity: 6.19 Mbq/mg; radiochemical purity: (12/00) 99.4%; repurified 2 times during study, (5/7/02) 99.2%, (5/17/02) 99.1%). Dosing preparations were supplemented with unlabeled S-1264RTZ (lot no. 010621G, purity: 99.3%). In the Excretion Study, three animals were dosed orally by gavage daily for 21 days with the test material and urine and fecal samples were

collected up daily through 21 days and at 24, 48, and 72 hours and 5, 7, 10 and 14 days post-final dose. In the Tissue Distribution Study, 12 animals were dosed orally by gavage with the test material and 3 animals/time point were euthanized after 10, 16, and 21 doses and 7 days post-final dose. In the Excretion Study, 75% of the total administered dose was excreted in the urine and 22% in the feces. The liver and kidneys were the primary sites of radiolabel recovery over the course of the dosing period with the level of radioactivity stabilizing over the dosing period. At 14 days post-final dose, the liver, red blood cell and hair were the primary locations where the radiolabel was recovered. The level of radioactivity recovered from the stomach, small intestine, cecum and large intestine was stabilized or slightly increased over the course of the dosing period. Radiolabel recovered in the hair peaked at the time of the 16st dose. The radiolabel was not sequestered in the fat nor did a significant fraction of the administered dose pass across the blood:brain barrier. Based upon the isolated metabolites, the major pathways of metabolism were cleavage of the ester linkage, oxidation of methyl groups on the benzyl moiety of the molecule to form alcohol, aldehyde and carboxylic acid metabolites, reduction of the double bond in the acid moiety of the molecule, and the formation of glutathione and sulfate adducts. **Study supplemental.** (Moore, 10/23/06)